

AMENDMENTS TO THE CLAIMS

1-22. (Canceled)

23. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:

- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate;
- scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one first objective, thereby receiving measuring values of optical parameters which are processed by means of signal processing for characterization of the at least one entity;
- the at least one entity substantially maintains its position with respect to the substrate for the duration of receiving the measuring values,
- generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and a second objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity;

- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic;
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection is used for measuring position of the interface and, thus, for indirectly positioning the measuring volume; and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner.

24. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:

- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,
- scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of

optical parameters which are processed by means of signal processing for characterization of the at least one entity,

- the at least one entity substantially maintains its position with respect to the substrate for the duration of the scanning,
- generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,
- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner,

wherein for indirectly positioning the measuring volume, the auxiliary focus relative to the interface is moved substantially along the optical axis of the objective generating the auxiliary focus, the intensity of the retroreflection dependent on movement of the

detector is registered and the position of the auxiliary focus is readjusted in a manner that the intensity of the retroreflection reaches its maximum.

25. (Previously Presented) Method according to claim 23 wherein the extension of the confocal detected volume of the auxiliary focus in the direction of the optical axis of the first objective is smaller than the extension of the measuring volume in said direction.
26. (Previously Presented) Method according to claim 25 wherein the auxiliary focus for obtaining a smaller extension of the confocal detected volume of the auxiliary focus is generated by a second objective having a numeric aperture which is larger than the numeric aperture of the first objective used for generating the measuring volume.
27. (Previously Presented) Method according to claim 25 wherein for obtaining the small extension of the confocal detected volume of the auxiliary focus a smaller one of the numerical aperture of a common optic or the respective first optic and second optic is used for generating the measuring volume than for generating the auxiliary focus.
28. (Previously Presented) Method according to claim 25 wherein for obtaining a small extension of the confocal detected volume of the auxiliary focus, a confocal arranged diaphragm is used at the detection of the auxiliary focus, said diaphragm having a smaller

opening than a confocal arranged diaphragm used at the detection of the measuring volume.

29. (Previously Presented) Method according to claim 23 wherein for indirectly positioning the measuring volume the auxiliary focus relative to the interface is moved substantially along the optical axis of the objective generating the auxiliary focus, the intensity of the retroreflection dependent on movement of the detector is registered and the position of the auxiliary focus is readjusted in a manner that the intensity of the retroreflection reaches its maximum.

30. (Previously Presented) Method according to claim 23 wherein for indirectly positioning the measuring volume the auxiliary focus relative to the interface is moved both, laterally to the optical axis of the optic generating the auxiliary focus and axially.

31. (Previously Presented) Method according to claim 29 wherein the amplitude of the periodical movement of the auxiliary focus is smaller than or equal to the extension of the measuring volume in the direction of the optical axis of the first objective.

32. (Previously Presented) Method according to claim 23 wherein the intensity of the retroreflection is detected by means of at least two detectors and the position of the

interface is determined by means of the distribution of the intensities detected by the detectors.

33. (Previously Presented) Method according to claim 23 wherein the scatter-light-intensity and/or the scatter-light-intensity dependent on the polarization and/or the fluorescence-intensity at at least one wavelength and/or the fluorescence-intensity in dependence on the polarization and/or the fluorescence-durability and/or molecular luminosity and/or Raman-scattering and/or luminescence are detected as optical parameters.
34. (Previously Presented) Method according to claim 23 wherein, in said step of arranging the at least one entity on and/or in a substrate, said substrate is a mineral or organic substrate chosen from the group consisting of a polymeric gel, a polymeric particle built up from inorganic material, a vesicular structure, a cell, a bacterium and a virus.
35. (Previously Presented) Method according to claim 23 wherein entities and/or substrates selected by means of the optical parameters are separated during or after the scanning process from the other entities and/or substrates.
36. (Currently Amended) The method according to claim 23, wherein said method for optically detecting ~~is applied to~~ further comprises a step of performing research of active ingredients, functional analysis of combinatoric-chemical or combinatoric-biological

synthesis-products, functional genome-analysis, evolutive biotechnology, diagnostics, proteom-analysis, or the investigation of material.

37. (Previously Presented) Apparatus for performing the method according to claim 23 for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria or virus, comprising:

- at least one first radiation source as well as at least one device being confocal or configured for multi-photon-excitation said device comprising a first objective and at least one first detector for detecting measuring values from the measuring volume;
- at least a second radiation source as well as at least one further device comprising a second objective and at least one second detector for detecting a retroreflection from an auxiliary focus, said second detector having a confocal arranged diaphragm, or a plurality of second detectors for detecting a retroreflection from an auxiliary focus, said second detectors having diaphragms arranged in front of and/or behind an image plane, and along the optical axis of the second objective generating the auxiliary focus;
- at least one device for positioning measuring volume and auxiliary focus relative to a substrate;
- a device for variably positioning the auxiliary focus relative to the measuring volume;
- a first optic collimating the radiation generated by the first radiation source; and

- a second optic being different from the first optic collimating the radiation generated by the second radiation source.
38. (Currently Amended) Apparatus for performing a method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:
- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,
 - scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of optical parameters which are processed by means of signal processing for characterization of the at least one entity,
 - the at least one entity substantially maintains its position with respect to the substrate for the duration of the ~~scanning~~ scanning,
 - generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,

- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner, said method optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic material, vesicular structures, cells, bacteria or virus, the apparatus having
- at least one first radiation source as well as at least one device being confocal or configured for multi-photon-excitation said device comprising an objective and at least one first detector for detecting measuring values from a measuring volume,
- at least one second radiation source as well as at least one further device comprising the same objective and a second detector for detecting a retroreflection from an auxiliary focus, the second detector having a confocal arranged diaphragm, or a plurality of second detectors for detecting a retroreflection from an auxiliary focus,

said second detectors having diaphragms arranged in front of and/or behind the image plane and along the optical axis of the objective generating the auxiliary focus,

- at least one device for positioning the measuring volume and auxiliary focus relative to the substrate,
- a device for relative positioning the auxiliary focus relative to the measuring volume,
- a first optic collimating the radiation generated by the first radiation source, and
- a second optic different from the first optic collimating the radiation generated by the second radiation source,

wherein the device for positioning the auxiliary focus relative to the measuring volume comprises means for variation of the convergence of bundles of rays that are focused by the respective objective for generation of the auxiliary focus and the measuring volume.

39. (Canceled)

40. (Previously Presented) Apparatus according to claim 37 wherein the device for positioning the measuring volume and the auxiliary focus relative to the substrate comprises means for positioning the auxiliary focus relative to the measuring volume.

41. (Previously Presented) Apparatus according to claim 37 wherein the device for positioning the auxiliary focus relative to the measuring volume comprises means for adjusting the relative position of the objectives to each other.
42. (Previously Presented) Apparatus according to claim 37 wherein the device for positioning the auxiliary focus relative to the measuring volume comprises means for variation of the convergence of bundles of rays that are focused by the respective objective for generation of the auxiliary focus and the measuring volume.
43. (Currently Amended) The apparatus according to claim 37, wherein said method for optically detecting ~~is applied to~~ further comprises a step of performing research of active ingredients, functional analysis of combinatoric-chemical or combinatoric-biological syntheses-products, functional genome-analysis, evolutive biotechnology, diagnostics, proteome-analysis, or the examining of material.
44. (Previously Presented) Method according to claim 23, wherein said substrate is arranged on a support.
45. (Previously Presented) Method according to claim 24, wherein said substrate is arranged on a support.

46. (Previously Presented) Method according to claim 29, wherein for indirectly positioning the measuring volume the auxiliary focus relative to the interface is moved periodically substantially along the optical axis of the objective generating the auxiliary focus.
47. (Previously Presented) Method according to claim 24 wherein the extension of the confocal detected volume of the auxiliary focus in the direction of the optical axis of the objective is smaller than the extension of the measuring volume in said direction.
48. (Previously Presented) Method according to claim 47 wherein for obtaining the small extension of the confocal detected volume of the auxiliary focus a smaller one of the numerical aperture of a common optic or the respective first optic and second optic is used for generating the measuring volume than for generating the auxiliary focus.
49. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:
- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,

- scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of optical parameters which are processed by means of signal processing for characterization of the at least one entity,
- the at least one entity substantially maintains its position with respect to the substrate for the duration of the scanning,
- generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,
- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner,

wherein the extension of the confocal detected volume of the auxiliary focus in the direction of the optical axis of the objective is smaller than the extension of the measuring volume in said direction,

wherein for obtaining the small extension of the confocal detected volume of the auxiliary focus a confocal arranged diaphragm is used at the detection of the auxiliary focus, said diaphragm having a smaller opening than a confocal arranged diaphragm used at the detection of the measuring volume.

50. (Canceled)

51. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:

- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,
- scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of

optical parameters which are processed by means of signal processing for characterization of the at least one entity,

- the at least one entity substantially maintains its position with respect to the substrate for the duration of the scanning,
- generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,
- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner,

wherein for indirectly positioning the measuring volume the auxiliary focus relative to the interface is moved both, laterally to the optical axis of the optic generating the auxiliary focus and axially.

52. (Previously Presented) Method according to claim 24 wherein the amplitude of the periodical movement of the auxiliary focus is smaller than or equal to the extension of the measuring volume in the direction of the optical axis of the objective.
53. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:
- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,
 - scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of optical parameters which are processed by means of signal processing for characterization of the at least one entity,
 - the at least one entity substantially maintains its position with respect to the substrate for the duration of the scanning,
 - generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least

partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,

- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner,

wherein the intensity of the retroreflection is detected by means of at least two detectors and the position of the interface is determined by means of the distribution of the intensities detected by the detectors.

54. (Previously Presented) Method according to claim 24 wherein the scatter-light-intensity and/or the scatter-light-intensity dependent on the polarization and/or the fluorescence-intensity at at least one wavelength and/or the fluorescence-intensity in dependence on the polarization and/or the fluorescence-durability and/or molecular luminosity and/or Raman-scattering and/or luminescence are detected as optical parameters.

55. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:

- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,
- scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of optical parameters which are processed by means of signal processing for characterization of the at least one entity,
- the at least one entity substantially maintains its position with respect to the substrate for the duration of the scanning,
- generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,

- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and
- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner,

wherein, in said step of arranging the at least one entity on and/or in a substrate, said substrate is a mineral or organic substrate chosen from the group consisting of a polymeric gel, a polymeric particle built up from inorganic material, a vesicular structure, a cell, a bacterium and a virus.

56. (Previously Presented) Method for optically detecting at least one entity chosen from the group consisting of molecules, molecule complexes, polymers, polymeric particles, particles built up from inorganic materials, vesicular structures, cells, bacteria and virus, comprising:

- arranging the at least one entity on and/or in a substrate, said substrate having a refraction-index which is different from the one of an at least one component adjacent to the substrate,
- scanning the at least one entity with a measuring volume using at least one device being confocal or configured for multi-photon-excitation, said device comprising a first radiation source and at least one objective, thereby receiving measuring values of optical parameters which are processed by means of signal processing for characterization of the at least one entity,
- the at least one entity substantially maintains its position with respect to the substrate for the duration of the scanning,
- generating, before and/or during the scanning step, an auxiliary focus by means of at least one second radiation source and the objective, said auxiliary focus is at least partly arranged on an interface between substrate and adjacent component or on another interface having a defined spacial relation to said entity,
- collimating the radiation generated by the first radiation source by a first optic and collimating the radiation generated by the second radiation source by a second optic being different from the first optic,
- detecting a retroreflection from the auxiliary focus by a detector having a confocal arranged diaphragm or by a plurality of detectors having diaphragms being arranged in front of and/or behind an image plane, and along the optical axis of the objective

generating the auxiliary focus, said retroreflection for measuring position of the interface and, thus, for indirectly positioning the measuring volume, and

- adjusting position of the auxiliary focus relative to the measuring volume in a defined manner,

wherein entities and/or substrates selected by means of the optical parameters are separated during or after the scanning process from the other entities and/or substrates.

57. (Currently Amended) The method according to claim 24, wherein said method for optically detecting ~~is applied to~~ further comprises a step of performing research of active ingredients, functional analysis of combinatoric-chemical or combinatoric-biological synthesis-products, functional genome-analysis, evolutive biotechnology, diagnostics, proteom-analysis, or the investigation of material.

58. (Previously Presented) Method according to claim 24, wherein for indirectly positioning the measuring volume the auxiliary focus relative to the interface is moved periodically substantially along the optical axis of the objective generating the auxiliary focus.

59. (Previously Presented) Apparatus according to claim 38 wherein the device for positioning the measuring volume and the auxiliary focus relative to the substrate comprises means for positioning the auxiliary focus relative to the measuring volume.

60. (Canceled)

61. (Currently Amended) The apparatus according to claim 38, wherein said method for optically detecting ~~is applied to~~ further comprises a step of performing research of active ingredients, functional analysis of combinatoric-chemical or combinatoric-biological syntheses-products, functional genome-analysis, evolutive biotechnology, diagnostics, proteome-analysis, or the examining of material.

62. (Previously Presented) Apparatus according to claim 39 wherein the device for positioning the measuring volume and the auxiliary focus relative to the substrate comprises means for positioning the auxiliary focus relative to the measuring volume.

63. (Canceled)

64. (Previously Presented) The apparatus according to claim 39, wherein said method for optically detecting is applied to research of active ingredients, functional analysis of combinatoric-chemical or combinatoric-biological syntheses-products, functional genome-analysis, evolutive biotechnology, diagnostics, proteome-analysis, or the examining of material.